

The Proliferative Tetrathyridium of *Mesocestoides vogae* sp. n. (Cestoda)

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ABSTRACT: The name *Mesocestoides vogae* sp. n. is proposed for the species of asexually proliferative tetrathyridium described by Specht and Voge (1965) and now in popular use in many laboratories in the world. A number of developmental differences are cited that make the tentative identification of this species as *Mesocestoides corti* Hoepli, 1925, by Specht and Voge difficult to accept. Until further data become available which demonstrate its adult stage and definitive host, a different name should be used for this metacestode species.

KEY WORDS: *Mesocestoides corti*, *Mesocestoides lineatus*, *Mesocestoides vogae* sp. n., asexual proliferation, taxonomy, Cestoda.

Mesocestoides corti was first named and described in the adult form by Hoepli (1925). Using a vial of about 100 preserved worms "collected by W. W. Cort in 1909 at Colorado Springs, Colorado" from "a specimen of the common house mouse, *Mus musculus*," Hoepli described these adult worms and named them in honor of the collector. Specht and Voge (1965) described a form of asexually proliferative tetrathyridia in the fence lizard, *Sceloporus occidentalis biseriatus*, and demonstrated the remarkable proliferative ability of these worms in the coelom and liver of laboratory mice. They tentatively identified this tetrathyridium as *M. corti*, with the disclaimer that "this might suggest our species is not *M. corti*" because they were not able to obtain development to a definitive size in either mice or cats. Voge (1967) further expressed her doubt of the identity of these larvae, stating that "Specht and Voge (1965) described asexual multiplication of tetrathyridia apparently belonging to *Mesocestoides corti*" (emphasis added by the writer). Beaver (1989) seriously questioned that mice could be the type host of *Mesocestoides corti*, because *M. corti* adults have never been rediscovered in the house mouse in over 60 ensuing years, nor has any experimental study shown that the proliferative tetrathyridium isolated by Specht and Voge (1965) can develop into an adult worm comparable in size to Hoepli's (1925) original description. Conn (1990) reviewed the literature concerning asexual reproduction in species of tetrathyridia and concluded that the only verified instance of this ability is seen in the strain isolated by Specht and Voge (1965). The present study is an attempt to resolve the question of the identity of this unique species of *Mesocestoides*.

Materials and Methods

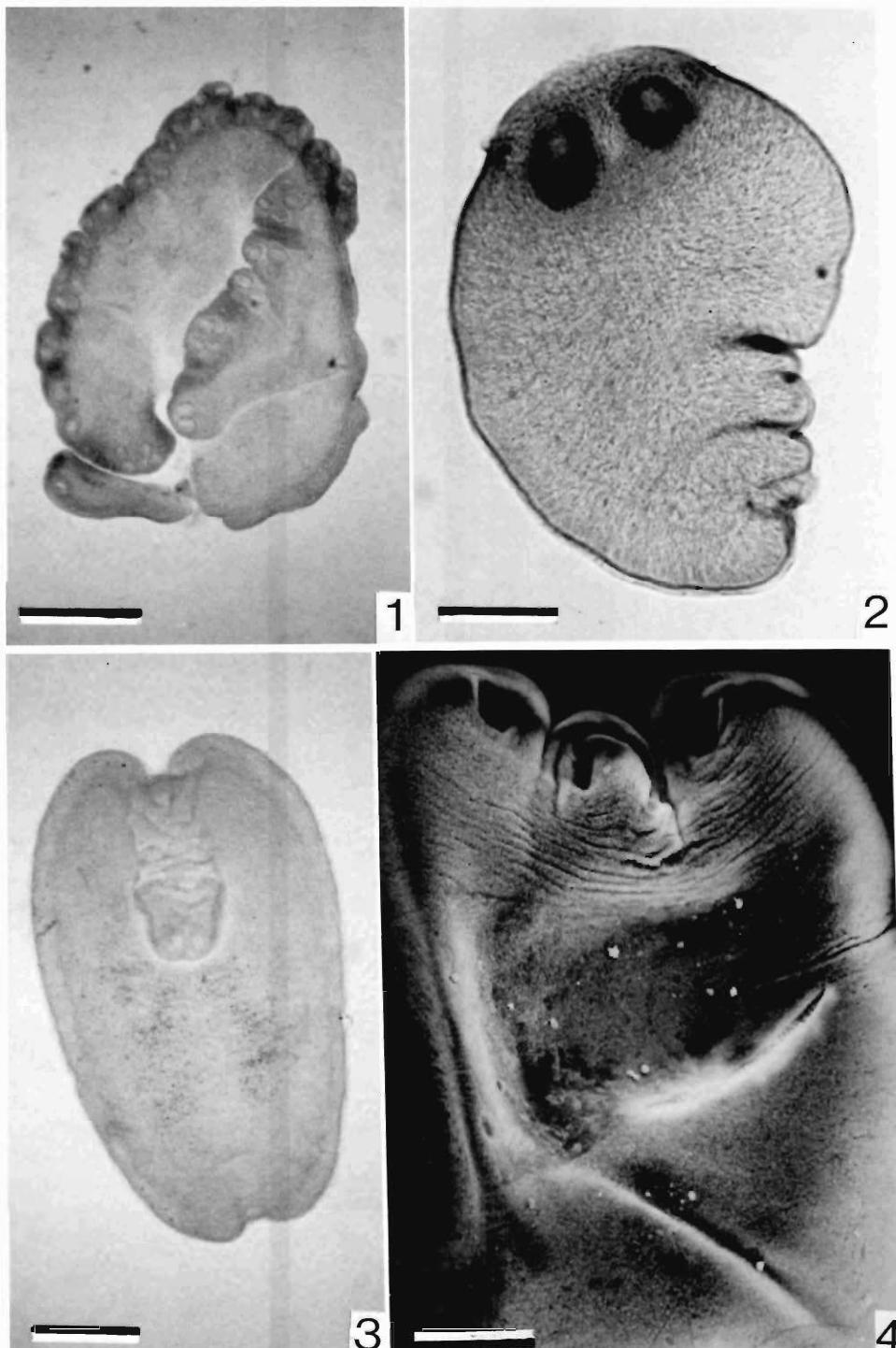
The asexually proliferative tetrathyridia used in this study were from a lineage originally obtained from the western fence swift by Specht and Voge (1965) and maintained in random bred BALB-C mice. About 150 worms obtained from the coelom of infected mice were introduced by stomach tube in all experimental infections. Non-proliferative tetrathyridia of *Mesocestoides lineatus* (Goeze, 1782) were obtained from *Anolis carolinensis* purchased from a commercial supplier as described by Conn and Etges (1984). Adults developed from these tetrathyridia in Syrian hamsters.

Microscope slide preparations were made of specimens fixed in neutral-buffered formalin and stained with Semichon's acetic acid carmine. When the non-specific esterase substrate bromindoxyacetate was used, specimens were fixed after 1-3-hr periods in the substrate solution (Thompson, 1966). Cover glass pressure was used only when needed for large specimens.

Specimens for scanning electron microscopy were fixed in modified Karnovsky's fixative, soaked overnight in 0.1 M cacodylate buffer, dehydrated in ethanol, critical point dried with CO₂, mounted on aluminum stubs, coated with 30 nm of gold/palladium in a sputter coater, and observed with an ETEC Autoscan electron microscope at 20 kV.

Results

In a period of 6 to 12 mo, mice were infected by many thousands of tetrathyridia actively proliferating by both multiple fission (Fig. 1) and binary fission (fissiparity) (Fig. 4). All growing and reproductive stages showed typical everted scolices (Fig. 2). Most tetrathyridia were found free in the coelom, but a considerable number were encapsulated in liver tissue and (rarely) in other organs such as mammary glands. Proliferative tetrathyridia from mice never showed any significant increase in size or ability to strobilize in the hamster intestine, but they were able to survive for more than 2 mo in this location, while undergoing asexual proliferation.



Figures 1–4. Tetrathyridia of *Mesocestoides vogae* (1, 2, 4) and *M. lineatus* (3). 1. Holotype specimen showing multiple scolex formation (Semichon's acetocarmine); scale bar = 0.25 mm. 2. Paratype specimen showing typical everted scolex (Bromindoxylin acetate stain); scale bar = 0.12 mm. 3. *M. lineatus*, a typical nonproliferative tetrathyridium with inverted scolex (Semichon's acetocarmine); scale bar = 0.50 mm. 4. SEM image showing binary scolex formation (fissiparity); scale bar = 0.25 mm.

Mesocestoides lineatus tetrathyridia never underwent reproduction when transplanted into the coelom of mice or hamsters, but remained the same in size and form as when recovered from the coelom of their anole hosts (Fig. 3). In the small intestine of 1 hamster, 3 gravid adult worms were obtained experimentally after only 22 days; these strobilae were 35, 165, and 208 mm long and were composed of about 100–400 proglottids.

Proliferative tetrathyridia from mice are the basis for the description of a new species.

Description

Mesocestoides vogae sp. n.

(Figs. 1, 2, 4)

DIAGNOSIS: Cestoda: Mesocestoidea. Metacestodes of the tetrathyridial type, ranging in size from 0.10 to 0.25 mm in length (Fig. 2); asexually proliferative by means of multiple (Fig. 1) and binary fission (fissiparity) (Fig. 4). Primarily situated in coelom and liver of reptiles (naturally) and small mammals (experimentally); occasionally able to invade other organs (of experimental hosts), such as mammary glands, mesenteries, testes, etc. Able to survive and replicate in small intestine of small mammals, elongate to 4–5 mm, but not strobilize. Occasionally able to strobilize slightly in experimental hosts such as cats and skunks, but rarely to fully gravid condition or length greater than 5 mm.

TYPE HOST: *Sceloporus occidentalis biseriatus* (Hallowell, 1854), the western fence lizard.

EXPERIMENTAL HOSTS: Mice, hamsters, cats, dogs, and other mammals.

TYPE LOCALITY: Riverside and Los Angeles counties, California.

ETYMOLOGY: The name is in honor of Marietta Voge, late Professor of Microbiology at the University of California at Los Angeles, who discovered this strain of proliferative tetrathyridia, and drew attention to it as a useful experimental model system.

SPECIMENS DEPOSITED: Holotype: USNM Helm. Coll. No. 81558. Paratypes: USNM Helm. Coll. No. 81559.

Discussion

Since its erection by Vaillant (1863), the genus *Mesocestoides* has remained enigmatic. The complete life cycle has never been demonstrated experimentally (though generally believed to involve 3 obligate hosts), the systematic relationships of the genus are uncertain, and the validity

of many named species is questionable because controlled experimental infections are not possible without knowing the identity of the first intermediate host, presumably an arthropod (Webster, 1949). Judging from the recent comprehensive account of Schmidt (1986), adult *Mesocestoides* are almost always parasitic in the small intestines of various carnivores, except for 2 species found in birds and the original report of *M. corti* in the house mouse by Hoeppli (1925). Mueller (1930) distinguished *M. corti* from *M. latus* Mueller, 1927, on the basis of the shorter length (4–8 cm), smaller number of proglottids (200–300), and smaller egg size (0.035 mm) in *M. corti*. Voge (1955) revised the genus, placing *M. variabilis* Mueller, 1927, and *M. manteri* Chandler, 1942, into synonymy with *M. corti* Hoeppli, 1925, based on her study of 3 preserved worms of the 100 used by Hoeppli (1925). She concluded that "I am reasonably certain that *Mesocestoides corti* Hoeppli and *Mesocestoides variabilis* Mueller are cospecific." Neither Hoeppli, Mueller, nor Voge commented on the presence of 100 adult worms in the intestine of a single house mouse.

Tetrathyridia have often been reported in host surveys, as has their development into various species of *Mesocestoides*. For example, Loos-Frank (1980) reported a typical case of adult development in cats experimentally exposed to tetrathyridia of *M. leptothylacus*, a natural parasite of foxes. She also showed that this species of tetrathyridium did not proliferate asexually in mice, birds, or voles. A similar result was obtained by the writer, using tetrathyridia from the common anole, *Anolis carolinensis*, reported by Conn and Etges (1984). In 1 Syrian hamster, 3 adult strobilae were obtained in only 22 days postinfection. These worms, tentatively identified as *M. lineatus* by Conn et al. (1984), were shown to survive indefinitely with no asexual proliferation as tetrathyridia (Fig. 3) in the coelom of mice and hamsters in several experiments, including the present report.

The first, and probably only, proven description of a proliferative tetrathyridium species is that of Specht and Voge (1965), who recovered their material from the coelom of the fence lizard, *Sceloporus occidentalis biseriatus*, in southern California. They demonstrated the remarkable ability of these worms to proliferate asexually, by a process termed "fissiparity," in vitro maintenance of the tetrathyridia for 4.5 mo, and the ease with which this parasite can be

transferred from mouse to mouse by stomach intubation. They also stressed the possibility of exploiting this convenient experimental model, a suggestion that has led to hundreds of publications produced by laboratories worldwide. All such studies descend from this 1 landmark report and the generosity of the discoverers who have provided this parasite for everyone's use. However, because Specht and Voge were unable to obtain adult worms in mice or cats, their identification of the species was very tentative; they suggested that "our species (might) not (be) *M. corti*. Voge (1967) expressed further uncertainty of its identity in her study, stating that Specht and Voge described asexual multiplication of tetrathyridia "apparently belonging to *Mesocestoides corti*." Uncertainty notwithstanding, the name *M. corti* is universally used to refer to this proliferative metacestode.

Several reports, mainly employing in vitro techniques (Voge, 1967; Eckert et al., 1969; Barrett et al., 1982; Kawamoto et al., 1986), have provided some evidence of growth (up to 1.84 cm), strobilization (up to 40 proglottids), and sexual maturation (a few embryonated eggs of about $\frac{1}{3}$ the diameter of those described for *M. corti* (0.035 mm) by Hoepli [1925]). Because the first intermediate host of *Mesocestoides* is yet unknown, infectivity of eggs produced by the stunted adults derived from proliferative tetrathyridia cannot be tested. It would be of some interest if they could be shown to hatch and develop in vitro in the manner described by Voge (1967) for eggs taken from adults found in natural infections, but there has been no report of this to the writer's knowledge. In any case, identification of proliferative tetrathyridia will remain as tentative as Voge believed, until they can be shown to produce adult worms comparable to the 4–8 cm worms described by Hoepli (1925).

The identity of the mouse as type host of *M. corti* was seriously questioned by Beaver (1989), because no confirmation of a natural infection in mice has been reported in over 60 yr. Furthermore, the presence of about 100 adult worms of 4–8-cm length in the intestine of so small a host is surprising, a point not commented upon by either Hoepli (1925), Mueller (1930), or Voge (1955). One can only speculate on the reasons that might explain this peculiarity in the description of *M. corti* and its host.

Conn's (1990) comprehensive literature review led to his conclusion that asexual proliferation is a proven ability in only 1 species of

tetrathyridium, i.e., that described by Specht and Voge (1965), and the suggestion that it "may be no more than a rare anomaly that has been propagated because of its convenience as a laboratory model." The writer concurs in these views, but suggests that perhaps asexual proliferation in *Mesocestoides* tetrathyridia may be a valuable evolutionary adaptation to the scarcity of definitive hosts, allowing for it to be transmitted directly from one vertebrate to another by deleting intermediate and definitive hosts from the normal life cycle. Confirmation of this view will depend on fieldwork to confirm the presence of asexual proliferation in a larger number of cases than presently known (1 fence lizard and 1 horned toad; Voge, pers. comm.).

Because of the several questions raised by Conn (1990) and Beaver (1989), and the uncertain identity of the proliferative tetrathyridium species discovered by Specht and Voge (1965), the writer is convinced that another name should be coined for it, and that *M. corti* be restricted to the adult worms described by Hoepli (1925) and any other morphologically similar adults found in any mammalian host. While Wittenberg (1934) cautioned against the naming of species of tetrathyridia because "no specific morphological distinction may be recognized between them" and "only in limited instances is it possible to indicate to what species they should be attributed," the writer thinks that there are good and sufficient developmental and physiological grounds (cited above) for giving a separate name to the proliferative tetrathyridia isolated by Specht and Voge.

Acknowledgments

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MEETING NOTICES

The Second International Symposium on Monogenea will be held in Montpellier, France, 5–8 July 1993. For information contact:

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The Twenty-first International Nematology Symposium will be held in Albufeira, Portugal, 12–17 April 1992. For information contact:

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